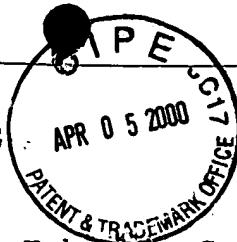


SEQUENCE LISTING



<110> Yano, Tetsuya; Nomoto, tsuyoshi; Immura, Takeshi; Canon Kabushiki Kaisha

<120> DNA Fragment Carrying Toluene Monooxygenase Gene,
Recombinant Plasmid, Transformed Microorganism,
Method for Degrading Chlorinated Aliphatic Hydrocarbon
Compounds and Aromatic Compounds, and
Method for Environmental Remediation

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<140> 09/430029

<141> 1999-10-29

<150> JP P1998-310801

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Met
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CONJ
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Leu Gly Ile Ala Leu
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cgc	gaa	acg	atc	gag	gaa	ctg	acc	ggc	acg	cgc	ttc	gac	ctg	cag	cag		1686		
Arg	Glu	Thr	Ile	Glu	Glu	Leu	Thr	Gly	Thr	Arg	Phe	Asp	Leu	Gln	Gln				
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Leu	Gln	Val	Asn	Leu	Ile	Thr	Leu	Ser	Gly	His	Ile	Asp	Glu	Asp	Asp			
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gac	gag	ttc	acg	ctg	agc	tgg	tcg	cac	tgaacgcccgc	gccacgcgc							1781	
Asp	Glu	Phe	Thr	Leu	Ser	Trp	Ser	His										
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Gly Thr Ala Ala Ile Asn Lys
1590 1595

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aactgcggtt cctgctgccc agcggccatg aactgcggct gttcgcgaag aaggcgctgg 5619
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ccgtgcactg gctcgaccac tgcctgctga tgtgcgaact gaacccggag gccggcgtga 5739
accgcgtcga ggagaacacg cgcttcatgg ccgagtgtct cgacttccat ctggccgagc 5799
aggtgatggt cggccgggc aacacgatc 5828

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Cont'

<210> 2
<211> 70
<212> PRT
<213> Burkholderia cepacia

<220>
<223> TomK polypeptide

<400> 2
Met Asn Gln His Pro Thr Asp Leu Ser Pro Phe Asp Pro Gly Arg Lys
1 5 10 15
Cys Val Arg Val Thr Gly Thr Asn Ala Arg Gly Phe Val Glu Phe Glu
20 25 30
Leu Ser Ile Gly Gly Ala Pro Glu Leu Cys Val Glu Leu Thr Leu Ser
35 40 45
Pro Ala Ala Phe Asp Ala Phe Cys Arg Glu Gln Gln Val Thr Arg Leu
50 55 60
Asp Val Glu Ala Asn Pro
65 70

<210> 3
<211> 331
<212> PRT
<213> Burkholderia cepacia

<220>
<223> TomL polypeptide

<400> 3
Met Thr Ile Glu Leu Lys Thr Val Asp Ile Lys Pro Leu Arg His Thr
1 5 10 15
Phe Ala His Val Ala Gln Asn Ile Gly Gly Asp Lys Thr Ala Thr Arg
20 25 30
Tyr Gln Glu Gly Met Met Gly Ala Gln Pro Gln Glu Asn Phe His Tyr
35 40 45
Arg Pro Thr Trp Asp Pro Asp Tyr Glu Ile Phe Asp Pro Ser Arg Ser
50 55 60
Ala Ile Arg Met Ala Asn Trp Tyr Ala Leu Lys Asp Pro Arg Gln Phe
65 70 75 80
Tyr Tyr Ala Ser Trp Ala Thr Thr Arg Ala Arg Gln Gln Asp Ala Met
85 90 95
Glu Ser Asn Phe Glu Phe Val Glu Ser Arg Arg Met Ile Gly Leu Met
100 105 110
Arg Asp Asp Val Ala Ala Arg Ala Leu Asp Val Leu Val Pro Leu Arg
115 120 125
His Ala Ala Trp Gly Ala Asn Met Asn Asn Ala Gln Ile Cys Ala Leu
130 135 140
Gly Tyr Gly Thr Val Phe Thr Ala Pro Ala Met Phe His Ala Met Asp
145 150 155 160
Asn Leu Gly Val Ala Gln Tyr Leu Thr Arg Leu Ala Leu Ala Met Ala
165 170 175
Glu Pro Asp Val Leu Glu Ala Ala Lys Ala Thr Trp Thr Arg Asp Ala
180 185 190
Ala Trp Gln Pro Leu Arg Arg Tyr Val Glu Asp Thr Leu Val Val Ala
195 200 205
Asp Pro Val Glu Leu Phe Ile Ala Gln Asn Leu Ala Leu Asp Gly Leu
210 215 220
Leu Tyr Pro Leu Val Tyr Asp Arg Phe Val Asp Glu Arg Ile Ala Leu
225 230 235 240
Glu Gly Gly Ser Ala Val Ala Met Leu Thr Ala Phe Met Pro Glu Trp
245 250 255
His Thr Glu Ser Asn Arg Trp Ile Asp Ala Val Val Lys Thr Met Ala
260 265 270

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Ay
Cont

Ala Glu Ser Asp Asp Asn Arg Ala Leu Leu Ala Arg Trp Thr Arg Asp
275 280 285
Trp Ser Ala Arg Ala Glu Ala Ala Leu Ala Pro Val Ala Ala Arg Ala
290 295 300
Leu Gln Asp Ala Gly Arg Ala Ala Leu Asp Glu Val Arg Glu Gln Phe
305 310 315 320
His Ala Arg Ala Ala Arg Leu Gly Ile Ala Leu
325 330

<210> 4
<211> 89
<212> PRT
<213> Burkholderia cepacia

<220>
<223> TomM polypeptide

<400> 4
Met Ser Asn Val Phe Ile Ala Phe Gln Ala Asn Glu Asp Ser Arg Pro
1 5 10 15
Ile Val Asp Ala Ile Val Ala Asp Asn Pro Arg Ala Val Val Val Glu
20 25 30
Ser Pro Gly Met Val Lys Ile Asp Ala Pro Asp Arg Leu Thr Ile Arg
35 40 45
Arg Glu Thr Ile Glu Glu Leu Thr Gly Thr Arg Phe Asp Leu Gln Gln
50 55 60
Leu Gln Val Asn Leu Ile Thr Leu Ser Gly His Ile Asp Glu Asp Asp
65 70 75 80
Asp Glu Phe Thr Leu Ser Trp Ser His
85

W Cont
<210> 5
<211> 516
<212> PRT
<213> Burkholderia cepacia

<220>
<223> TomN polypeptide

<400> 5
Met Asp Thr Pro Thr Leu Lys Lys Lys Leu Gly Leu Lys Asp Arg Tyr
1 5 10 15

Ala Ala Met Thr Arg Gly Leu Gly Trp Glu Thr Thr Tyr Gln Pro Met
 20 25 30
 Asp Lys Val Phe Pro Tyr Asp Arg Tyr Glu Gly Ile Lys Ile His Asp
 35 40 45
 Trp Asp Lys Trp Val Asp Pro Phe Arg Leu Thr Met Asp Ala Tyr Trp
 50 55 60
 Lys Tyr Gln Gly Glu Lys Glu Lys Leu Tyr Ala Val Ile Asp Ala
 65 70 75 80
 Phe Thr Gln Asn Asn Ala Phe Leu Gly Val Ser Asp Ala Arg Tyr Ile
 85 90 95
 Asn Ala Leu Lys Leu Phe Leu Gln Gly Val Thr Pro Leu Glu Tyr Leu
 100 105 110
 Ala His Arg Gly Phe Ala His Val Gly Arg His Phe Thr Gly Glu Gly
 115 120 125
 Ala Arg Ile Ala Cys Gln Met Gln Ser Ile Asp Glu Leu Arg His Tyr
 130 135 140
 Gln Thr Glu Thr His Ala Met Ser Thr Tyr Asn Lys Phe Phe Asn Gly
 145 150 155 160
 Phe His His Ser Asn Gln Trp Phe Asp Arg Val Trp Tyr Leu Ser Val
 165 170 175
 Pro Lys Ser Phe Phe Glu Asp Ala Tyr Ser Ser Gly Pro Phe Glu Phe
 180 185 190
 Leu Thr Ala Val Ser Phe Ser Phe Glu Tyr Val Leu Thr Asn Leu Leu
 195 200 205
 Phe Val Pro Phe Met Ser Gly Ala Ala Tyr Asn Gly Asp Met Ser Thr
 210 215 220
 Val Thr Phe Gly Phe Ser Ala Gln Ser Asp Glu Ser Arg His Met Thr
 225 230 235 240
 Leu Gly Ile Glu Cys Ile Lys Phe Leu Leu Glu Gln Asp Pro Asp Asn
 245 250 255
 Val Pro Ile Val Gln Arg Trp Ile Asp Lys Trp Phe Trp Arg Gly Tyr
 260 265 270
 Arg Leu Leu Thr Leu Val Ala Met Met Met Asp Tyr Met Gln Pro Lys
 275 280 285
 Arg Val Met Ser Trp Arg Glu Ser Trp Glu Met Tyr Ala Glu Gln Asn
 290 295 300
 Gly Gly Ala Leu Phe Lys Asp Leu Ala Arg Tyr Gly Ile Arg Glu Pro
 305 310 315 320
 Lys Gly Trp Gln Asp Ala Cys Glu Gly Lys Asp His Ile Ser His Gln
 325 330 335
 Ala Trp Ser Thr Phe Tyr Gly Phe Asn Ala Ala Ser Ala Phe His Thr
 340 345 350
 Trp Val Pro Thr Glu Asp Glu Met Gly Trp Leu Ser Ala Lys Tyr Pro
 355 360 365

Asp Ser Phe Asp Arg Tyr Tyr Arg Pro Arg Phe Asp His Trp Gly Glu
370 375 380
Gln Ala Arg Ala Gly Asn Arg Phe Tyr Met Lys Thr Leu Pro Met Leu
385 390 395 400
Cys Gln Thr Cys Gln Ile Pro Met Leu Phe Thr Glu Pro Gly Asn Pro
405 410 415
Thr Lys Ile Gly Ala Arg Glu Ser Asn Tyr Leu Gly Asn Lys Phe His
420 425 430
Phe Cys Ser Asp His Cys Lys Asp Ile Phe Asp His Glu Pro Gln Lys
435 440 445
Tyr Val Gln Ala Trp Leu Pro Val His Gln Ile His Gln Gly Asn Cys
450 455 460
Phe Pro Pro Asp Ala Asp Pro Gly Ala Glu Gly Phe Asp Pro Leu Ala
465 470 475 480
Ala Val Leu Asp Tyr Tyr Ala Val Thr Met Gly Arg Asp Asn Leu Asp
485 490 495
Phe Asp Gly Ser Glu Asp Gln Lys Asn Phe Ala Ala Trp Arg Gly Gln
500 505 510
Ala Thr Arg Asn
515

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t

<210> 6
<211> 118
<212> PRT
<213> Burkholderia cepacia

<220>
<223> TomO polypeptide

<400> 6
Met Ala Val Ile Ala Leu Lys Pro Tyr Asp Phe Pro Val Lys Asp Ala
1 5 10 15
Val Glu Lys Phe Pro Ala Pro Leu Leu Tyr Val Cys Trp Glu Asn His
20 25 30
Leu Met Phe Pro Ala Pro Phe Cys Leu Pro Leu Pro Pro Asp Met Pro
35 40 45
Phe Gly Ala Leu Ala Gly Asp Val Leu Pro Pro Val Tyr Gly Tyr His
50 55 60
Pro Asp Phe Ala Lys Ile Asp Trp Asp Arg Val Glu Trp Phe Arg Ser
65 70 75 80
Gly Glu Pro Trp Ala Pro Asp Pro Ala Lys Ser Leu Ala Gly Asn Gly
85 90 95
Leu Gly His Lys Asp Leu Ile Ser Phe Arg Thr Pro Gly Leu Asp Gly

100 105 110

Leu Gly Gly Ala Ser Phe
115

<210> 7
<211> 354
<212> PRT
<213> Burkholderia cepacia

<220>
<223> TomP polypeptide

<400> 7

Met Ser His Gln Leu Thr Ile Glu Pro Leu Gly Val Thr Ile Glu Val
1 5 10 15
Glu Glu Gly Gln Thr Met Leu Asp Ala Ala Leu Arg Gln Gly Ile Tyr
20 25 30
Ile Pro His Ala Cys Cys His Gly Leu Cys Gly Thr Cys Lys Val Ala
35 40 45
Val Leu Asp Gly Glu Thr Asp Pro Gly Asp Ala Asn Pro Phe Ala Leu
50 55 60
Met Asp Phe Glu Arg Glu Glu Gly Lys Ala Leu Ala Cys Cys Ala Thr
65 70 75 80
Leu Gln Ala Asp Thr Val Ile Glu Ala Asp Val Asp Glu Glu Pro Asp
85 90 95
Ala Glu Ile Ile Pro Val Arg Asp Phe Ala Ala Asp Val Thr Arg Ile
100 105 110
Glu Gln Leu Thr Pro Thr Ile Lys Ser Ile Arg Leu Lys Leu Ser Gln
115 120 125
Pro Ile Arg Phe Gln Ala Gly Gln Tyr Val Gln Leu Glu Ile Pro Gly
130 135 140
Leu Gly Gln Ser Arg Ala Phe Ser Ile Ala Asn Ala Pro Ala Asp Val
145 150 155 160
Ala Ala Thr Gly Glu Ile Glu Leu Asn Val Arg Gln Val Pro Gly Gly
165 170 175
Leu Gly Thr Gly Tyr Leu His Glu Gln Leu Ala Thr Gly Glu Arg Val
180 185 190
Arg Leu Ser Gly Pro Tyr Gly Arg Phe Phe Val Arg Arg Ser Ala Ala
195 200 205
Arg Pro Met Ile Phe Met Ala Gly Gly Ser Gly Leu Ser Ser Pro Arg
210 215 220
Ser Met Ile Ala Asp Leu Leu Ala Ser Gly Val Thr Ala Pro Ile Thr
225 230 235 240

Leu Val Tyr Gly Gln Arg Ser Ala Gln Glu Leu Tyr Tyr His Asp Glu
245 250 255
Phe Arg Ala Leu Ala Glu Arg His Pro Asn Phe Thr Tyr Val Pro Ala
260 265 270
Leu Ser Glu Gly Ala Pro His Ala Gly Gly Asp Val Ala Gln Gly Phe
275 280 285
Val His Asp Val Ala Lys Ala His Phe Gly Gly Asp Phe Ser Gly His
290 295 300
Gln Ala Tyr Leu Cys Gly Pro Pro Ala Met Ile Asp Ala Cys Ile Thr
305 310 315 320
Thr Leu Met Gln Gly Arg Leu Phe Glu Arg Asp Ile Tyr His Glu Lys
325 330 335
Phe Ile Ser Ala Ala Asp Ala Gln Gln Thr Arg Ser Pro Leu Phe Arg
340 345 350
Arg Val

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WT
<210> 8
<211> 118
<212> PRT
<213> Burkholderia cepacia

<220>
<223> TomQ polypeptide

<400> 8
Met Asp Ala Gly Arg Val Cys Gly Thr Val Thr Ile Ala Gln Thr Asp
1 5 10 15
Glu Arg Tyr Ala Cys Val Ser Gly Glu Ser Leu Leu Ala Gly Met Ala
20 25 30
Lys Leu Gly Arg Arg Gly Ile Pro Val Gly Cys Leu Asn Gly Gly Cys
35 40 45
Gly Val Cys Lys Val Arg Val Leu Arg Gly Ala Val Arg Lys Leu Gly
50 55 60
Pro Ile Ser Arg Ala His Val Ser Ala Glu Glu Asn Asp Gly Tyr
65 70 75 80
Ala Leu Ala Cys Arg Val Val Pro Asp Gly Asp Val Glu Leu Glu Val
85 90 95
Ala Gly Arg Leu Arg Lys Pro Phe Phe Cys Gly Met Ala Cys Ala Gly
100 105 110
Thr Ala Ala Ile Asn Lys
115

<210> 9
<211> 36
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Designed PCR primer

<400> 9
agtccgccat ggaggcgaca ccgatcatga atcagc 36

<210> 10
<211> 34
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Designed PCR primer

<400> 10
caccgaccat ggatcagcac cccaccgatc tttc 34

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<210> 11
<211> 34
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Designed PCR primer

<400> 11
tgccgccttc catgggttct gccgcgaaca gcag 34

<210> 12
<211> 39
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Designed PCR primer

<400> 12
agcaagccat ggccatcgag ctgaagacag tcgacatca 39

<210> 13
<211> 35
<212> DNA
<213> Artificial Sequence
<220>
<223> Description of Artificial Sequence: Designed PCR primer

*a
Cont*
<400> 13
ccgaccatca cctgctcggc cagatgaaag tcgag 35